Innovations in Agriculture 2023, 6:01-05
www.innovationsagriculture.com

ISSN: 2617-1295
DOI: https://doi.org/10.25081/ia.2023-026

Research Article

Effect of Black Soldier Fly (*Hermetia illucens*) larvae feed supplement on length-weight relationship of Nile Tilapia (*Oreochromis niloticus*)

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ARTICLE INFO

Received: October 03, 2023
Revised: November 02, 2023
Accepted: November 02, 2023
Published: November 02, 2023

Keywords:

Black soldier fly; feed supplement; length-weight relationship; nile tilapia

ABSTRACT

One of the main challenges in the aquaculture sector is the high cost of fishmeal, which accounts for about 70 percent of production costs. The black soldier fly (BSF) larvae meal could be an alternative because of its high protein and essential fatty acids contents that are important for fish to grow and develop normally. An analysis was carried out to examine the effect of various feeding strategies on the growth and performance of Nile tilapia (Oreochromis niloticus). Five diets with equal nitrogen content were enriched with BSF larvae meal at different proportions (0%, 25%, 50%, 75%, and 100%). The meal with 0% BSF was used as a control. The length and weight of the fish were collected biweekly, while mortality data were collected daily. The study assessed the length and weight relationship of the fish using the parameter 'b'(regression coefficient). From this research, 'b' was found to range from 3.230 to 3.635, indicating isometric growth patterns. The well-being of the experimental fish was also evaluated through the condition factor 'K,' which ranged from 0.907 to 1.002, suggesting that all the fish were in good health. We, therefore, conclude that substituting the conventional fish means with black soldier fly larvae appears to be possible and impacts fish growth normally.

INTRODUCTION

The Nile tilapia (Oreochromis niloticus) is an omnivorous and herbivorous freshwater cichlid. It mainly consumes phytoplankton, macrophytes, insects, litter, phytoplankton algae, and plant material [1]. In addition, it is highly resistant to harsh environmental conditions, consuming both formulated diets and natural foods, which make it economically viable [2]. The intricate relationship between the size and weight of fish species, known as the length-weight relationship, is a fundamental aspect of fisheries and aquaculture research [3]. The length-weight relationship in Nile tilapia has been extensively explored as a tool for assessing the state of health and well-being of individuals and populations [4, 5, 6, 7]. A positive correlation between length and weight indicates healthy growth, while deviations from this relationship may signify changes in feeding habits, environmental conditions, or other stressors [8]. Such studies have been integral in assessing the success of aquaculture operations and fisheries management.

One of the main challenges in the aquaculture sector is the high cost of fishmeal, which accounts for about 70 percent of production costs [9]. The black soldier fly (BSF) larvae meal could be an alternative because of its high protein content [2]. Additionally, BSF larvae have essential fatty acids that are important for fish to grow and develop normally [10]. For instance, silver cyprinid fish (Rastrineobola argentea) contains about 40% crude protein [11], whereas BSF crude protein levels typically range from 28 to 48%, and lipid levels range from 12 to 42% [12]. They are more nutritious than traditional fish meal products. This strategy is environmentally and economically sustainable, as it can produce nutrient-dense food from organic waste, acting as a recycling machine with little environmental harm. Furthermore, BSFs are more ecologically friendly alternatives to aquaculture since they use less water and area.

Studies have examined the length-weight relationship to gain insights into growth dynamics and health conditions. For example, studies by Ahmed et al. [5] and Agboola et al. [7] investigated the length-weight relationship of Nile tilapia in various aquatic environments, providing valuable information about the species' growth and adaptation patterns. These investigations have underscored the importance of understanding the length-weight relationship as a critical indicator of fish health and aquaculture success. Despite the existing body of research, there remains a gap in understanding how alternative dietary supplements, such as black soldier fly larvae meal, might influence the length-weight relationship of Nile tilapia. Therefore, this study aimed to address this gap by exploring the effects of varying levels of black soldier fly larvae supplementation on the length-weight relationship of Nile tilapia. By extending the knowledge in this field, this research contributes to a more comprehensive understanding of how different dietary components interact with growth patterns and overall health in this economically significant fish species.

MATERIALS AND METHODS

The research was conducted in a laboratory at the School of Agriculture and Food Sciences, Jaramogi Oginga Odinga University of

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Science and Technology (JOOUST), Bondo, Kenya. Mono sex male O. niloticus fingerlings weighing 15±0.06 grams were obtained from Lake Agro Limited in Siaya. The fingerlings were transported in specialized containers and allowed to acclimatize to laboratory conditions in aquarium containers for 14 days before the start of the experiment. The experiment employed a completely randomized design (CRD) with five treatments: Commercial fish meal (Control) (0% BSF), BSF2 (25%), BSF3 (50%), BSF4 (75%), and BSF5 (100%). The treatments had a crude protein content of 29-30%. Each treatment was replicated three times. The research work was undertaken in 90-liter capacity rectangular glass aquaria of dimensions 70 x 35 x 40cm each. Each aquarium contained 15 fingerlings. All fingerlings were fed experimental diets three times a day, at three-hour intervals from morning to evening throughout the study period. The feed was given at 3% of their body weight daily. The daily feed ratio was divided equally among all treatments on each feeding schedule. The study was conducted for over 12 weeks under laboratory conditions.

Ethical considerations

The research sought the approval of the Ethical Review Committee (ERC), JOOUST Postgraduate School, and the National Commission for Science, Technology, and Innovation (NACOSTI) before the beginning of the study. The study also considered ethical guidelines for using fish in experimental designs [1]. Healthy fish stocks were starved a day prior to their transportation to the JOOUST Lab. Fish were transported in oxygenated fish bags the following day early morning to curb stress to the fish. The lab water used in the aquaria was left to stand for two days to dechlorinate before use in the glass aquaria. During the study period, the welfare of the experimental fish was maintained by ensuring they were not stressed and healthy.

Formulated diets

Five isonitrogenous approximately (30%) and isocaloric approximately 18 ME/Kg/DM test diets were formulated using an Excel solver linear programming model (Table 1). The diets were formulated with ingredients such as *H. illucens* larva meal (BSF), soya bean meal, maize, wheat pollard, wheat bran, sunflower meal, sunflower oil, fishmeal, salt, di-calcium phosphate (DCP) and vitamin premixes. The diets were ground and hand-mixed, then pelleted using standard commercial screen sizes of 2mm, after which they were cooled and packaged. Pelleting of formulated fishmeal diets was done at Great Lakes Feed Company in Bondo. After diet formulation, diets were also analyzed according to AOAC [13] to determine the nutritional value and varying crude protein levels. The diets were isonitrogenous, approximately 30% crude protein (Table 2).

Data collection procedure

Data on temperature, water quality, ammonia level, pH, and growth performance (weight and length) parameters were collected. Water quality parameters were frequently monitored during the experiment to provide suitable conditions for optimal fish growth performance. During the experiment, an adequate dissolved oxygen level was maintained through aeration using an aquarium bubbler. The experimental tank water temperature, pH, and dissolved oxygen (DO) were measured three times daily, at 9:00 a.m., 1:00 p.m., and 4:00 p.m., and recorded in a water quality parameter worksheet. Dissolved oxygen was measured using an oxygen meter (Yellow Springs, YSI 550A U.S.A). Temperature and pH were measured using an electronic pH probe.

Using a plastic filter basket, fish were removed from each aquarium, and a digital weighing balance was used to weigh the fish to the closest 0.01g. Length (in centimeters) was measured using a fish measuring board. The mean biweekly growth was computed using the weight and length values. The specific growth rate mean weight gain and length gain were computed using the biweekly data. Mortality rates each day were noted. Growth performance metrics, including final mean weight, mean weight gain, and specific growth rate, were computed at the end of the trial using weight and length data.

Growth analysis

Linear regression analysis was used to determine the length-weight (log-transformed) relationships, and length-weight scatter diagrams were plotted. The length-weight relationship of the fish was calculated using the cube law [14].

i) $W = aL^b$
Where W = fish weight (g), L = Total length (cm), a = regression intercept and b = regression slope. The logarithmic transformation for the formula is;
$$\text{Log } W = \log a + b \log L$$
When the values of 'b' equal 3, the growth is classified as isometric; otherwise, it is classifiedas allometric.

ii) Fulton's Condition factor (K) determined the health index of the fish
$$K = 100 \frac{W}{L^b}$$
Where K = Condition factor, L = Total length of fish in centimetres (cm), b = Regression Co-efficient obtained from length-weight equation.

Table 1. Inclusion levels of feed ingredients in *O. niloticus* diets.

Ingredients	FM	BSF2	BSF3	BSF4	BSF5
%Inclusion	**0%**	**25%**	**50%**	**75%**	**100%**
BSF	0.00	4.15	8.30	12.45	16.60
Maize	10.00	10.00	10.00	10.00	10.00
Soya meal	33.50	33.50	33.50	33.50	33.50
Wheat bran	14.00	14.00	14.00	14.00	14.00
Wheat pollard	20.00	20.00	20.00	20.00	20.00
Sunflower cake	5.00	5.00	5.00	5.00	5.00
Sunflower oil	0.10	0.10	0.10	0.10	0.10
Fishmeal	16.60	12.45	8.30	4.15	0.00
Di-calcium phosphate	0.10	0.10	0.10	0.10	0.10
Rock salt	0.20	0.20	0.20	0.20	0.20
Vitamin premix	0.50	0.50	0.50	0.50	0.50

Table 2. Proximate analysis of formulated diets.

	Diets				
Chemical %composition	**FM (0%)**	**BSF2 (25%)**	**BSF3 (50%)**	**BSF4 (75%)**	**BSF5 (100%)**
Dry Matter (%)	89.0	89.5	89.8	89.8	89.0
Moisture (%)	11.0	10.5	10.2	10.2	11.0
Crude protein (%)	31.8	29.2	29.6	30.4	30.6
Crude Fibre (%)	4.5	4.0	6.0	6.5	5.5
Ether Extract (%)	6.4	7.6	6.8	8.0	8.4

RESULTS AND DISCUSSIONS

The physicochemical properties of all the aquarium water did not differ significantly from one another. Table 3 displays the means and standard errors of the water quality metrics. The measurements of the physicochemical water quality indicators showed only minor differences across all treatments. BSF3 had the lowest mean pH of 6.96, whereas BSF4 had the highest mean pH of 7.23. The mean temperature that was measured was 23.24°C at the lowest and 24.00°C at the highest points. At 4.31 mg/L, the lowest oxygen levels were found in BSF3, while at 4.91 mg/L, the highest levels were found in BSF4. BSF4 had the greatest levels of ammonia, measuring 0.12 mg/L; the values for the other treatments were essentially identical.

All treatments had high regression coefficient values ranging between 0.98 and 0.99 were obtained from the length-weight relationship (Table 4). In this study, the mean value 'b' ranged from 3.23 to 3.63. BSF4, BSF5, and FM (3.63, 3.49, and 3.44, respectively) had a higher regression coefficient (b) than BSF2 and BSF3 (3.25, 3.23). The range of Fulton's condition factor K ranged from 0.9 to 1.0. BSF2 had a condition factor of 1.00, while BSF3 and FM had K values of 0.99 and 0.98, respectively. K values were 0.92 for BSF5 and 0.90 for BSF4. BSF4 had lower condition factors than FM, BSF2, and BSF3. The log-transformed data fit well in the linear model (Fig 1) for all diets, with R2 values above 0.9. Positive isometric growth was observed in all treatment diets.

BSF4 diets had a higher value of positive isometric growth (b= 3.63) than the control, FM diet (b= 3.44), and the other three diets, BSF2 (b= 3.23), BSF3 (b= 3.33), and BSF5 (b= 3.49), indicating that fish become more rotund as their lengths increase. O. niloticus fed FM and diets supplemented with H.illucens BSF2, BSF3, BSF4, and BSF5 all had isometric growth, indicating that they become heavier as they increase in length.

The high coefficient of determination 'R' values in each treatment indicated that these O. niloticus equations were relatively accurate. Fulton's condition factor K had a range of 0.90 to 1.00. The condition factor for BSF2 was 1.00, while the condition factors for BSF3 and FM were 0.99 and 0.98, respectively. BSF5 (0.92) and BSF4 (0.90) had lower condition factors than FM, BSF2, and BSF3. The length-weight relationship's findings were consistent with those of Anani and Nunno's [15] study, which demonstrated isometric growth (b≥ 3) growth in Nile tilapia fed with a farm-made tilapia diet. The findings of this study contrast with those of Mugendi et al. [16], who discovered that O.noliticus fed commercial diets supplemented with duckweed (Lemna minor) initially exhibited allometric growth before switching to isometric growth in the sixth week. For all treatments, the condition factor (K) was approximately 1 (Table 4). The diet supplemented with H.illucens at 25% had the highest condition factor of all treatments. These findings suggest that O.niloticus was in good health throughout the experiment. Despite having higher isometric growth than O.niloticus fed FM, BSF2, BSF3, and BSF5, O.niloticus supplemented with H.illucens at 75% had the lowest condition factor (0.9) compared to the other experimental fish. As evidenced by the fish's low final weight, the low condition factor in O. niloticus fed diets supplemented with H. illucens at 75% indicates that it had poor feed intake compared to the others. According to Bagenal [8], the fish condition factor also reveals a species' level of feeding activity to demonstrate whether it effectively utilizes its feeding resource. Based on the condition factor of BSF2 (K=1) and (b=3.2), O.niloticus thrives more on diets supplemented with *H. illucens* at 25% compared to other levels of supplementation. Fish with a higher condition factor (above 0.9) are typically healthy and thriving. In contrast, a lower condition factor (below 0.9) might suggest that the fish is underweight or malnourished.

While all of the fish in the current study were in good health and condition, the condition factor registered was lower than that of Luo et al. [17], who reported a higher condition factor above 3 in all diets when *O. niloticus* was fed diets supplemented with betaine in aquaria. The results could be attributed to higher water quality parameters observed compared to the current study. Anani and Nunno [15] also registered condition factors above 1.00 when *O. niloticus* was fed commercial and farm-made diets. The reported condition factor is closely related to Shahabuddin et al. [18], who reported condition factors of about 1.00 when juvenile *O. niloticus* were fed diets with *Pyropia spheroplasts*.

Since all of the fish in the experiment were male, their condition factors were nearly identical and the K ≥0.9 range. Male fish have larger bodies and more flesh than females [19]. The results of the length-weight relationship demonstrated that the *O. niloticus* were in good health when fed diets supplemented with *H. illucens* larva. The current study finding supports that O. niloticus can be reared with diets supplemented with *H. illucens* larva meal without harming their physiology. The results of this study support Cren's [14] finding that a fish's shape or body outline typically changes throughout its life. Studies on the length-weight relationship and condition factor of *O. niloticus* fed diets supplemented with *H. illucens* are scarce.

Table 3. Mean values of physicochemical parameters of water measured in aquaria (values are expressed as mean ± SEM).

Parameters	**FM**	**BSF2**	**BSF3**	**BSF4**	**BSF5**
%	**0%**	**25%**	**50%**	**75%**	**100%**
Temp (OC)	23.46±0.2201	24.01±0.2835	23.24±0.1970	23.66±0.1914	23.44±0.2434
NH3(Mg-L)	0.02±0.0003	0.02±0.0004	0.02±0.0009	0.12±0.0526	0.02±0010
NH4+(Mg-L)	3.75±0.7219	3.36±0.1501	3.40±0.1562	3.58±0.0433	3.14±0.2166
Oxygen(Mg-L)	4.83±0.2849	4.56±0.2425	4.31±0.2548	4.92±0.2960	4.91±0.2902
PH	7.14±0.1118	7.14±0.0909	6.99±0.1262	7.23±0.0751	6.95±0.1224

Table 4. Length-weight relationship equation parameters and condition factor of *O. niloticus*.

Diet	R	r2	A	b	k
FM	0.987	0.973	0.007	3.445	0.994
BSF2	0.990	0.981	0.010	3.230	1.002
BSF3	0.990	0.980	0.009	3.326	0.980
BSF4	0.992	0.984	0.005	3.635	0.907
BSF5	0.985	0.971	0.006	3.498	0.926

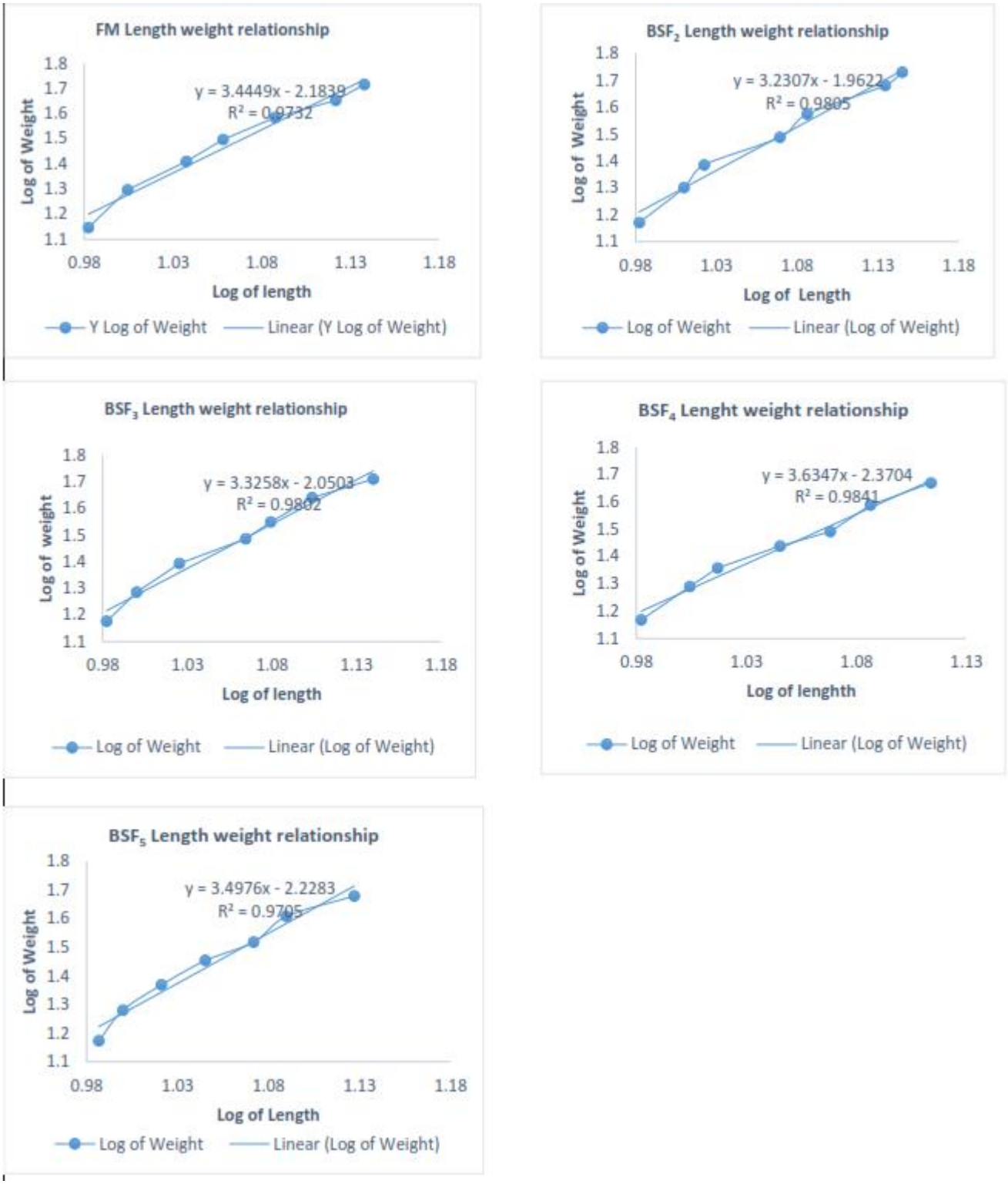

Fig. 1. Graph of the length-weight relationships of *O. niloticus* and prediction models by treatments.

CONCLUSION AND RECOMMENDATIONS

The findings suggest that incorporating black soldier fly larvae meal as a dietary supplement is feasible and can influence the growth dynamics and overall health of Nile tilapia. The observed variations in the length-weight relationship and condition factors emphasize the importance of diet composition in shaping the fish's growth trajectories and physiological state.

RECOMMENDATIONS

The study has identified and recommends the following critical areas for future research in aquaculture practices:
1. Further investigations should explore the optimal dietary proportions of black soldier fly larvae meal that would yield the most favorable length-weight relationship and condition factors. Fine-tuning the dietary composition can contribute to maximizing growth while maintaining fish health.
2. Assessing the economic feasibility of incorporating black soldier fly meal into commercial aqua feed formulations to determine the cost-effectiveness of this alternative feed source.

ACKNOWLEDGMENTS

We appreciate the Jaramogi Oginga Odinga University for supporting this study through the World Bank and collaborating with the Africa Center of Excellence in the Sustainable Use of Insects as Food and Feed (INSEFOODS).

CONFLICT OF INTEREST

The authors declare that there is no conflict of interests regarding the publication of this paper.

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